

**Western Riverside County  
Multiple Species Habitat Conservation Plan (MSHCP)  
Biological Monitoring Program**

**Arroyo Toad  
(*Anaxyrus californicus*)  
Survey Report 2010**



**23 March 2011**

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**NOTE TO READER:**

This report is an account of survey activities conducted by the Biological Monitoring Program for the Western Riverside County Multiple Species Habitat Conservation Plan (MSHCP). The MSHCP was permitted in June 2004. The Monitoring Program monitors the distribution and status of the 146 Covered Species within the Conservation Area to provide information to Permittees, land managers, the public, and the Wildlife Agencies (i.e., the California Department of Fish and Game and the U.S. Fish and Wildlife Service). Monitoring Program activities are guided by the MSHCP species objectives for each Covered Species, the information needs identified in MSHCP Section 5.3 or elsewhere in the document, and the information needs of the Permittees.

MSHCP reserve assembly is ongoing and it is expected to take 20 or more years to assemble the final Conservation Area. The Conservation Area includes lands acquired for conservation under the terms of the MSHCP and other lands that have conservation value in the Plan Area (called public or quasi-public lands in the MSHCP). In this report, the term “Conservation Area” refers to the Conservation Area as understood by the Monitoring Program at the time the surveys were planned and conducted.

We would like to thank and acknowledge the land managers in the MSHCP Plan Area, who in the interest of conservation and stewardship facilitate Monitoring Program activities on the lands for which they are responsible. A list of the lands where data collection activities were conducted in 2010 is included in Section 7.0 of the Western Riverside County Regional Conservation Authority (RCA) Annual Report to the Wildlife Agencies. Partnering organizations and individuals contributing data to our projects are acknowledged in the text of appropriate reports.

While we have made every effort to accurately represent our data and results, it should be recognized that data management and analysis are ongoing activities. Any reader wishing to make further use of the information or data provided in this report should contact the Monitoring Program to ensure that they have access to the best available or most current data.

The primary preparer of this report was the 2010 Arroyo Toad Project Lead, Jonathan Reinig. If there are any questions about the information provided in this report, please contact the Monitoring Program Administrator. If you have questions about the MSHCP, please contact the Executive Director of the RCA. Further information on the MSHCP and the RCA can be found at [www.wrc-rca.org](http://www.wrc-rca.org).

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## INTRODUCTION

Four amphibian species covered by the Western Riverside County MSHCP inhabit stream environments in southern California: arroyo toad (*Anaxyrus californicus*, formally *Bufo californicus*), California newt (*Taricha torosa*, formally coast range newt *T. torosa torosa*), California red-legged frog (*Rana draytonii*), and Sierra Madre yellow-legged frog (*Rana muscosa*). The Biological Monitoring Program has been collaborating with the Western Ecological Research Center, U.S. Geological Survey (USGS) and U.S. Forest Service (USFS) in conducting amphibian-stream surveys since 2004 to reduce overlapping survey efforts and to ensure consistent data collection methods. The purpose of the stream surveys has been to assess habitat suitability and to document breeding locations for arroyo toad, red-legged frog, California newt, and yellow-legged frog within species-specific Core Areas and/or their tributaries. We targeted different species from year to year, depending on efforts of collaborating agencies and reporting requirement for species-specific objectives. The target species for stream surveys in 2010 was arroyo toad.

Arroyo toad has narrow habitat requirements, typically being restricted to the middle reaches of third order streams below 3000 ft. in elevation (Stebbins 1951; Dudek & Associates 2003). They are additionally constrained by not inhabiting areas with a landscape slope of  $\geq 3\%$  or areas which lack sandy substrates (Miller and Miller 1936; Sweet 1992). Specific breeding habitat requirements limit the potential areas they are likely to inhabit (Stebbins 1951; Sweet 1989, 1992). Records of arroyo toad within the MSHCP Plan Area date from the mid 1930s to early 2000s (Dudek & Associates 2003). The historical locations for arroyo toad in western Riverside County include: Temecula Creek, Arroyo Seco Creek, Los Alamos Creek, San Jacinto River, Bautista Creek, Wilson Creek, Vail Lake (Core Areas), Tenaja Creek, and Cole Creek (non-core areas) (Dudek and Associates 2003).

The species objectives for arroyo toad require the conservation of 9 Core Areas in the MSHCP Conservation Area; 1) San Juan Creek, 2) Los Alamos Creek, 3) San Jacinto River, 4) Indian Creek, 5) Bautista Creek, 6) Wilson Creek, 7) Temecula Creek, 8) Arroyo Seco, and 9) Vail Lake. MSHCP species-specific objective 6 for arroyo toad states that within the MSHCP Conservation Area, Reserve Managers will maintain breeding populations at a minimum of 80 percent of the conserved breeding locations as measured by the presence/absence of juvenile toads, tadpoles, or egg masses across any 5 consecutive years (Dudek and Associates 2003).

Biological Monitoring Program biologists conducted stream surveys targeting arroyo toad in 2003 and each year from 2005-2008 (see Table 1 in Results section below). Past surveys included both areas where arroyo toad was known to recently occur and areas of possible occurrence based on presence of key habitat requirements. During these surveys, breeding populations of arroyo toad were documented in 3 of the 9 Core Areas: Arroyo Seco, Bautista Creek, and San Juan Creek. An additional breeding population was also found in Cole Creek on the Santa Rosa Plateau in 2005 (see *Western Riverside County MSHCP Biological Monitoring Program Arroyo Toad Survey Report 2005*). The Indian Creek and Vail Lake Core Areas had not been surveyed prior to 2010

because of lack of access to suitable habitat within these cores. In 2010, Vail Lake was not surveyed due to the above mentioned constraints, but portions of Indian Creek were accessed and surveyed.

In 2010, we primarily focused our surveys in Core Areas where arroyo toad had not been previously detected or surveyed for by the Biological Monitoring Program. This included portions of Los Alamos, Indian, Temecula, and Wilson Creeks and the San Jacinto River. We surveyed areas known to be recently occupied as well, when time and personnel allowed (i.e., San Juan Creek, Bautista Creek, and Arroyo Seco). Specifically, the overall survey goals and objectives for stream surveys in 2010 were as follows:

### **Goals and Objectives**

1. Collect information about arroyo toad distribution and demographics in Core Areas and other locations with potentially suitable habitat.
  - a. Survey for arroyo toad in as many areas as possible where historical populations existed and/or where suitable habitat is present.
2. Document and characterize potential breeding habitat of arroyo toad across Core Areas.
  - a. Assess presence/absence of key habitat components necessary for arroyo toad breeding.
  - b. Measure water quality and vegetation types (upland and riparian habitat) wherever flowing water occurs.
3. Collaborate with USGS on an ongoing population study of reptiles in southern California.
  - a. Retrieve tissue samples from USGS target species for genetic analysis and deliver them to USGS.

### **METHODS**

In 2005 we began conducting stream surveys following the *USGS Aquatic Species and Habitat Assessment Protocol for Southcoast Ecoregion Rivers, Streams, and Creeks* (USGS 2005). In general, the USGS protocol describes a visual encounter and dip-net survey method for detecting all life stages of amphibians, and includes an assessment of habitat characteristics. Using Hawth's Tools extension (Beyer 2004) in ArcGIS v.9.3 (ESRI 2006), we divided streams into 250-m reaches within the targeted drainages, and labeled them following the naming convention described in the USGS protocol. In areas without a known population of arroyo toad, or areas that had not been surveyed prior to 2010, formal surveys were usually preceded by assessment surveys. These assessment surveys were used to determine the presence/absence of water and to evaluate the suitability of the habitat for arroyo toads (see below).

We modified the USGS protocol in 2009 to better address MSHCP species-specific objectives for the target species. These modifications include using the Sawyer and Keeler-Wolf (1995) vegetation classifications for characterizing the surrounding landscape, collecting additional water chemistry data, recording weather data at the end

of the survey, and altering and adding characteristics recorded when species were observed. Furthermore, we began recording animal abundance, and the number of shallow, medium, and deep pools as continuous rather than categorical values. For the 2010 surveys, we added an arroyo toad habitat suitability assessment (USGS 2003) to help identify areas most suitable for arroyo toads. This habitat suitability assessment, originally developed by the USGS, asked for the presence/absence of 3 key habitat characteristics with which arroyo toad are usually associated: sandy aquatic substrate, sand bars caused by stream braiding, and sandy terraces adjacent to the stream channel. If all 3 habitat characteristics were present in a given stream reach, that reach was labeled “Good”. If only 2 were present it was labeled “Marginal”. If only 1, then it was labeled “Poor” arroyo toad habitat.

Tissue samples were also collected from USGS target species following a standardized protocol developed by Biological Monitoring Program staff (Appendix A).

### **Personnel and Training**

Training consisted of in-field mock stream surveys, as well as the study of identification guides of amphibian species likely to be encountered during arroyo toad surveys. The in-field portion of this training consisted of group, mock surveys in a known arroyo toad breeding area (San Juan Creek, May 24, 2010). During this training session, participants learned how to identify all life stages of most stream breeding amphibians likely to be encountered during the 2010 surveys, with the exception of western toad (*Anaxyrus boreas*), and bullfrog (*Lithobates catesbeianus*) which were not present during the training session. This in-field training also served as a method to familiarize surveyors with the Monitoring Program Stream Survey Protocol.

Biological Monitoring Program personnel were funded by the California Department of Fish and Game or the Regional Conservation Authority; volunteers are noted. The following staff conducted stream surveys in 2010:

- Jonathan Reinig (Project Lead, Biological Monitoring Program)
- Masanori Abe (Biological Monitoring Program)
- Melody Aimar (Volunteer, Santa Ana Watershed Association)
- Elizabeth Dionne (Biological Monitoring Program)
- Joanna Gibson (Biological Monitoring Program)
- Julie Golla (Biological Monitoring Program)
- Tara Graham (Biological Monitoring Program)
- Robert Packard (Biological Monitoring Program)
- Michael Robinson (Biological Monitoring Program)
- Esperanza Sandoval (Biological Monitoring Program)
- Sloane Seferyn (Biological Monitoring Program)
- Joseph Sherrock (Biological Monitoring Program)

### **Survey Site Selection**

Arroyo toad is constrained by inhabiting a very narrow habitat type, so we focused our surveys in areas containing essential habitat characteristics. Third order

streams were predominantly surveyed, although some reaches in second and fourth order streams were surveyed when other necessary habitat components were present. We excluded all stream reaches higher than 3,200 ft. above sea level, because arroyo toads are not found above this elevation (Stebbins 1951). We focused on stream reaches with a slope  $\leq 3\%$ , although some steeper reaches were included when they were surrounded by reaches with  $< 3\%$  slope gradient. Finally, we further limited our searches by only surveying in arroyo toad Core Areas or in tributaries immediately upstream from arroyo toad Core Areas. Once assessment surveys were under way, priority was given to formally survey stream reaches with “Good” and “Marginal” arroyo toad habitat types over areas characterized as “Poor”, using the USGS habitat suitability assessment described below (USGS 2005).

### **Survey Methods**

We conducted assessment surveys to locate the presence of water and to assess suitability of the habitat for arroyo toad before conducting more rigorous visual-encounter and dip-net surveys based on the USGS (2005) protocol. We did not conduct assessment surveys at San Juan Creek, Los Alamos Creek, Arroyo Seco, Bautista Creek, or portions of the San Jacinto River for presence of water, because previous MSHCP stream surveys have shown water to be flowing perennially and/or good arroyo toad habitat to be present at these locations. We deemed streams not appropriate for more rigorous visual-encounter and dip-net surveys if they were completely dry or would likely be dry within a few days of the assessment survey.

We began assessment surveys for suitable habitat on 28 April 2010 and conducted visual-encounter and dip-net surveys from 24 May to 14 July along 250-m reaches that contained water. All surveys were conducted in daylight hours (0800 – 1700) from downstream to upstream areas, along stream banks and within channels. We collected the following information on paper datasheets at the beginning and end of each surveyed reach: time, sky condition (0 = clear or few clouds, 1 = party cloudy or variable, 2 = cloudy or overcast, 3 = fog, 4 = mist or drizzle, 5 = showers or light rain, 6 = heavy rain, 7 = sleet or hail, 8 = snow), temperature ( $^{\circ}\text{C}$ ) in shade at 1 m above ground, average and max wind speeds (km/h), and presence/absence of water. At the beginning of each reach we recorded the date, observers, water temperature ( $^{\circ}\text{C}$ ), water transparency (clear, moderate/translucent, opaque), pH, dissolved oxygen (concentration [mg/L] and percent), conductivity (mS/cm), salinity (ppt), total dissolved solids (g/L), wetted width (m) and depth (m) of stream channel, water velocity (m/s), and number of wetted channel braids. We also took photos facing upstream at the start of each reach.

At the end of each reach, we recorded the presence/absence of the 3 principal habitat characteristics for arroyo toad described above. We also recorded presence and abundance of exotic plant species, percent wet length of the reach, number of each pool type (shallow, medium, and deep), presence and number of plunge pools, presence and type of aquatic refugia, bank full width (m), flood prone width (m), percent of overhead canopy throughout the reach, presence and type of basking areas, percent cover of the 3 most dominant riparian plants, percent cover of the 3 most common aquatic substrates, percent cover of the 3 most common bank substrates, upland and riparian community types, and presence, type, and level of recent disturbance. Furthermore, we calculated the

slope of each reach back in the Monitoring Program office using a Microsoft Excel function and the elevation of the start and end points.

We recorded each Covered Species encountered, recorded data for a cluster if multiple individuals were within the same approximately 10-m diameter area. The following information was recorded for Covered Species: location name, number, location coordinates, species, abundance (i.e., cluster size), age (adult, tadpole, egg mass), detection method (visual or audio), number of photos taken, photo ID number, tissue sample number (if applicable), and any relevant notes.

We recorded presence of non-covered species from each age class (e.g., tadpole, juvenile, egg mass) encountered per reach, and recorded the abundance along each reach. We totaled the estimated number of each age class for non-covered species at the end of the stream survey once the entire reach had been surveyed (e.g., 0 egg masses, 12 western toad tadpoles, ~50 California tree frog tadpoles, 5 western toad juveniles, 0 adults). All information listed above for Covered Species was recorded for non-covered species except location coordinates which were deemed unnecessary given we have start and end location coordinates for all stream reaches surveyed.

## RESULTS

We conducted habitat assessments and/or surveys for arroyo toad in 9 streams in 2010, including 8 of 9 Core Areas (Table 1). We confirmed occupancy and breeding in 3 Core Areas in 2010 (San Juan Creek, Bautista Creek, and Arroyo Seco).

**Table 1.** Arroyo toad survey locations and occupied locations from 2003 – 2010. Stream names in bold are arroyo toad Core Areas; shading denotes that surveys were conducted; ‘X’ denotes that arroyo toads were detected.

Stream Name	2003	2004	2005	2006	2007	2008	2009	2010
Arroyo del Toro	-	-		-	-	-	-	-
<b>Arroyo Seco</b>	X	-	X			-	-	X
<b>Bautista Creek</b>	X	-	X		-	-	-	X
Cole Creek	-	-	X			-	-	-
<b>Indian Creek</b>	-	-	-	-	-	-	-	
<b>Los Alamos Creek</b>	-	-		-		-	-	
Potrero Creek	-	-			-	-	-	
<b>San Jacinto River</b>		-		-	-		-	
<b>San Juan Creek</b>		-	X			-	-	X
Santa Gertrudis Creek	-	-		-	-	-	-	-
<b>Temecula Creek</b>	-	-		-	-	-	-	-
Tenaja Creek	-	-		-	-	-	-	-
Warm Springs Creek	-	-		-	-	-	-	-
<b>Wilson Creek</b>	-	-		-	-	-	-	
<b>Vail Lake</b>	-	-	-	-	-	-	-	-



All reaches were dry during pre-survey habitat assessments in the Temecula Creek Core Area; therefore no formal surveys were conducting in this core in 2010. The Vail Lake Core Area was not surveyed in 2010 due to lack of conserved land in the area.

We assessed sixty-five 250-m stream reaches in 2010. Only 9 of these reaches were deemed appropriate arroyo toad habitat and were wet enough to follow up with formal surveys. The remaining 56 reaches, which were assessed but not surveyed, were either dry (30 reaches, 53%), were categorized as “Poor” arroyo toad habitat (23 reaches, 42%) or were not surveyed due to time constraints (3 reaches, 5%).

We conducted surveys for arroyo toad in 79 stream reaches in 2010. Survey time per reach varied from 20 to 172 min, depending on streambed characteristics (e.g., slope, vegetation, rock barriers) and presence and abundance of amphibians detected (mean = 58.7 min). All reaches surveyed contained at least some water. We detected arroyo toads in a total of 13 reaches in the 3 occupied Core Areas (Figure 1, Appendix B). We did not observe arroyo toad in any previously unknown location.

Of the 13 reaches where arroyo toad was detected, 10 reaches were classified as “Good” arroyo toad habitat and 3 reaches were classified as “Marginal” arroyo toad habitat. Of the remaining 130 reaches assessed and/or surveyed, 65 were classified as “Good”, 19 were classified as “Marginal”, and 45 were classified as “Poor” arroyo toad habitat (Appendix B). A brief description of results by specific drainages appears below.

### **Temecula Creek**

Only 7 reaches in this creek and one of its immediate tributaries (Tule Creek) were assessed. All reaches were classified as “Good” arroyo toad habitat, but all reaches were dry at the time of the assessment. Therefore, no formal surveys were conducted in the Temecula Creek Core Area in 2010.

### **San Juan Creek**

We surveyed 19 stream reaches in San Juan Creek and its immediate tributaries (Morrell Creek, Long Creek, and an unnamed tributary). We detected arroyo toad in 6 of these reaches (Table 2). Prior to surveying, on 26 April 2010, a pair of adult arroyo toads were incidentally observed while in amplexus, and pools containing arroyo toad tadpoles were incidentally found in an additional 2 reaches. Therefore, we confirmed occupancy and breeding within this Core Area in 2010. San Juan Creek reaches were predominantly classified as “Good” and “Marginal” arroyo toad habitat.

### **Los Alamos Creek**

We surveyed 10 reaches in this Core Area. Despite an abundance of “Good” and “Marginal” arroyo toad habitat and despite it being located adjacent to San Juan Creek, we detected no evidence of arroyo toad in 2010.



**Table 2.** Arroyo toad detections in 2010.

Waterway	Stream		Tadpoles*	Metamorphs*	Juveniles	Adults
	Reach	Date				
Arroyo Seco	ARRS13	6/7/2010	20	-	-	-
Arroyo Seco	ARRS14	6/7/2010	1	-	-	-
Arroyo Seco	ARRS15	6/7/2010	31	-	-	-
Bautista Creek	BAUS54	6/10/2010	120	6	-	-
Bautista Creek	BAUS55	6/10/2010	26	1	-	-
Bautista Creek	BAUS58	6/10/2010	70	-	-	-
Bautista Creek	BAUS59	6/10/2010	3	-	-	-
San Juan Creek	SJUSS6	6/25/2010	1	-	-	-
San Juan Creek	SJUSS7	6/28/2010	-	4	4	-
San Juan Creek	SJUSS11	5/24/2010	450	-	-	-
Morrell Creek (trib of San Juan Creek)	MORS1	5/24/2010	1000	-	-	-
Morrell Creek (trib of San Juan Creek)	MORS2	7/2/2010	51	1	-	-
Morrell Creek (trib of San Juan Creek)	MORS5	7/2/2010	44	2	-	-
San Juan Creek trib 3**	SJUT3S1	4/26/2010	-	-	-	2
San Juan Creek trib 3**	SJUT3S2	4/26/2010	1000	-	-	-

\* numbers estimated; not individually counted.

\*\* incidental observations not made during surveys.

### San Jacinto River

With 42 reaches surveyed or assessed, the San Jacinto River and portions of its tributaries (Potrero, Sand, Strawberry, and Toll Road Creeks) was the most extensively searched area in 2010. Although the San Jacinto River has an abundance of “Good” and “Marginal” arroyo toad habitat and is located adjacent to a stream with a breeding population of arroyo toad (Bautista Creek), no evidence of arroyo toad was detected here in 2010.

### Arroyo Seco

We surveyed only 4 stream reaches in Arroyo Seco. All reaches were “Good” arroyo toad habitat. The remaining reaches of Arroyo Seco were classified as “Poor” habitat or contained none of the arroyo toad preferred habitat characteristics. Arroyo toad tadpoles were found in 3 reaches in Arroyo Seco, confirming occupancy of this Core Area in 2010, and indicating that breeding is occurring. Its immediate tributaries,

although limited in number, were not assessed or surveyed due to time constraints.

### **Indian Creek**

We assessed or surveyed 22 reaches in Indian Creek and 1 of its immediate tributaries (Mellor Creek). The majority of these (16 reaches) were classified as “Poor” arroyo toad habitat. There were no arroyo toads detected in this area in 2010.

### **Bautista Creek**

We surveyed or assessed 18 reaches in Bautista Creek. We detected arroyo toad tadpoles and/or metamorphs in 4 reaches confirming occupancy of this Core Area in 2010, and indicating that breeding is occurring. Six of the stream reaches were dry at the time they were surveyed or assessed. Most of the portion of Bautista Creek we surveyed contains “Good” arroyo toad habitat (Figure 2).

### **Wilson Creek**

We assessed 15 reaches but only 4 reaches were surveyed in Wilson Creek and its tributaries (Cahuilla Creek and an unnamed tributary). About half (7 reaches) of the reaches assessed were classified as “Good” arroyo toad habitat, but were dry. Less than half (6 reaches), contained water, but were classified as “Poor” arroyo toad habitat. We detected no arroyo toads in Wilson Creek or its tributaries.

### **Vail Lake**

Vail Lake was not assessed or surveyed because of lack of inclusion in the Conservation Area.

### **Cole Creek and Tenaja Creek**

Cole Creek and Tenaja Creek are the only other streams in the Plan Area known to historically contain arroyo toad. We did not survey these streams in 2010 because of time constraints and because they are not arroyo toad Core Areas. Arroyo toads have not been seen in Cole Creek since 2005 (*Carole Bell, The Nature Conservancy, personal communication*).

We detected many other Covered and non-covered species during arroyo toad surveys in 2010 (Appendix B). Covered Species observed included: California newt (*Taricha torosa*), orange-throated whiptail, (*Aspidoscelis hyperythra*; formerly Belding’s orange-throated whiptail, *A. h. beldingi*), coastal western whiptail (*Aspidoscelis tigris stejnegeri*; formerly *A. t. multiscutatus*), granite spiny lizard *Sceloporus orcutti*, granite night lizard (*Xantusia henshawi*), and arroyo chub (*Gila orcuttii*).

During arroyo toad surveys in 2010 Monitoring Program biologists collected tissue samples from 6 individuals of 2 species and delivered these samples to USGS in support of their ongoing population genetics study of reptiles in southern California (Appendix A).

## **DISCUSSION**

Our survey goals for 2010 were to document breeding arroyo toad in as many Core Areas as possible, and to assess the suitability of habitats available to this species



across the Conservation Area. Although we pointedly conducted numerous surveys outside of areas known to be recently occupied, we detected breeding evidence of arroyo toad solely in the same 3 streams that were known to be inhabited from previous Monitoring Program surveys (San Juan Creek, Bautista Creek, and Arroyo Seco). We did not find arroyo toad in any new locations or in any additional Core Areas in 2010.

The USGS habitat classification system was useful in screening out stream reaches that were unsuitable for arroyo toad. Arroyo toad was only detected in habitat classified as “Good” or “Marginal” (Appendix B). However, many reaches that were characterized as “Good” (n=65) or “Marginal” (n=19) habitat remained unoccupied.

Of the 3 Core Areas where arroyo toad breeding was confirmed, San Juan Creek had the largest population (based on number of occupied reaches and the total number of all life stages), Bautista Creek had the second largest, and Arroyo Seco had the smallest population at the time of our surveys. It is possible that additional populations of arroyo toad may be found in the immediate tributaries of San Juan Creek where there is appropriate habitat. Much of Bautista Creek was surveyed relatively late in the year when its upper reaches were drying up. These upper reaches may have contained arroyo toads earlier in the year, though we did not detect them during surveys in 2010. Immediate tributaries of Bautista Creek might also support arroyo toad where appropriate habitat exists. The reach of Arroyo Seco downstream from the surveyed reaches is likely inhabited by arroyo toad, but it is currently not in the Conservation Area and is therefore inaccessible to the Monitoring Program. Portions of 3 of Arroyo Seco’s unnamed tributaries, which are currently accessible could be inhabited by arroyo toad, but were not surveyed this year due to time constraints.

We may have failed to detect arroyo toad due to various factors in a few of the Core Areas in 2010. The majority of arroyo toad habitat in Temecula Creek is currently not in the Conservation Area. If populations still exist in this stream, they would likely be centered around Vail Lake. In this area there are reliable historical records of arroyo toad, and it is also near where Arroyo Seco converges with Temecula Creek, a known breeding location. This is approximately 6.5 stream miles downstream from the area where we surveyed. The portions of Indian Creek that were surveyed were dominated by “Poor” arroyo toad habitat. The lower reaches of Indian Creek, just above its confluence with the San Jacinto River, appear to have much better habitat for arroyo toad (low stream gradient, an abundance of sand, etc.) but are not currently in the Conservation Area. In Wilson Creek, the lower reaches have the best habitat for arroyo toads, and there are historical records of arroyo toads here. It is also near the breeding population at Arroyo Seco, but this area also is currently not in the Conservation Area.

The San Jacinto River and Los Alamos Creek, although classified as “Good” arroyo toad habitat in many reaches, and being located adjacent to streams with evidence of breeding arroyo toad, did not have any arroyo toad detections in 2010. The reason for their absence in these areas is unknown.

### **Recommendations for Future Surveys**

It is possible that arroyo toad still exist at Vail Lake. As such, it is an area of interest to the Biological Monitoring Program. There is historical observation data from

Vail Lake where 3 streams that contain, or are believed to contain breeding populations of arroyo toad converge: Wilson Creek, Temecula Creeks, and Arroyo Seco. The habitat around Vail Lake also visually appears to have suitable breeding habitat for arroyo toad when viewed from a nearby public road.

When personnel are available, surveys in the immediate tributaries of occupied streams within Core Areas would provide a better understanding of arroyo toad's distribution. Cole Creek should be thoroughly surveyed for arroyo toad to establish the distribution and relative population size. These surveys should be centered on the Clinton Keith Road overpass of Cole Creek where evidence of breeding arroyo toad was detected in 2005.

In areas where "Good" arroyo toad habitat was located, but that were dry at the time of our surveys, surveys should be conducted when water is present (earlier in the year or following a wetter winter). During assessment surveys, additional information such as vegetation community type, and any animal observations should be gathered. This will give a more complete summary of areas searched, as opposed to only collecting these data where formal surveys were conducted.

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[USGS] United States Geological Survey. 2005. Draft USGS Aquatic species and habitat assessment protocol for southcoast ecoregion rivers, streams, and creeks. Western Ecological Research Center. Sacramento, CA.



## **Appendix A. Western Riverside County MSHCP Biological Monitoring Program Protocol for Reptile Tissue Sampling, March 2009**

Tissue sampling has been shown to be a valuable component of scientific and genetic studies. Many genetic studies have revealed important results about local populations (Richmond and Jockusch 2007; Wood, Fisher, and Reeder 2007), and tissue sampling allows for analyses of population genetics to be conducted without killing individuals in the population. Reptiles generally recover quickly from injuries sustained during acquisition of a small tissue sample, and the resulting scars can be used to aid in recapture identification analysis. Scale clipping and taking tail tips rarely draws blood, and the application of a tissue adhesive (e.g., New Skin) will speed the healing process and stem any blood loss. The tissue adhesive should also help minimize the risk of bacterial infection, although this is a possible deleterious side-effect. Some species of lizards also readily shed their tails as a defense mechanism, and although care will be taken to process all animals as quickly and carefully as possible, it is likely that a small number of individuals will lose their tails during handling. Although there are some risks associated with tissue sampling, this method should have less impact on target populations than taking specimens for vouchering, and will still provide valuable monitoring data.

The protocol outlined below will be followed by Monitoring Program staff processing reptiles in the field. All current herpetological personnel were trained in taking tissue samples by a USGS biologist at the USGS office in San Diego on March 5, 2009, or trained by those who attended said training. All crew took tissue samples from dead specimens; however, we used a live specimen for demonstrating handling techniques while taking tissue samples. We will train future personnel on live specimens in the field. We will temporarily store all tissue samples in refrigeration at the MSHCP's Biological Monitoring Office at 4500 Glenwood Drive, Riverside, CA, and then transport them to the USGS Western Ecological Research Center's San Diego Field Office at 4165 Spruance Road, San Diego, CA for genetic analysis.

### **USGS TARGET SPECIES Processing Methods**

Target Species include: Gilbert's skink (*Plestiodon gilberti*), western skink (*P. skiltonianus*), rosy boa (*Lichanura trivirgata*), southern rubber boa (*Charina umbratica*), glossy snake (*Arizona occidentalis*), shovel-nosed snake (*Chionactis occipitalis*), San Diego mountain kingsnake (*Lampropeltis zonata pulchra*), San Bernardino mountain kingsnake (*L. z. parvirubra*), red coachwhip (*Masticophis flagellum*), striped whipsnake

(*M. lateralis*), red-sided garter snake (*Thamnophis sirtalis infernalis*), two-striped garter snake (*T. hammondi*), southwestern blind snake (*Leptotyphlops humilis humilis*) San Diego banded gecko (*Coleonyx variegatus abbotti*), western banded gecko (*C. v. variegatus*), granite night lizard (*Xantusia henshawi henshawi*), and sagebrush lizard (*Sceloporus vandenburgianus*).

The following information will be collected for each USGS Target Species.

1. Gender/Age
  - Male, female or unknown
2. Measurements
  - Using metric ruler
    - i. Snout-Vent length (mm)
    - ii. Tail length (mm)
  - Using pesola scale
    - i. Weight (g): tare scale first with sampling bag, then place animal in bag.
      1. Use the smallest scale possible for the most accuracy.
3. Take tissue sample (y/n) (Do not take a sample if the animal is too small to safely do so)
  - i. Label micro-centrifuge tubes with sample # [date, full board name(site#-board#), 4-letter species code, and individual sequential # (ex. 20091125\_MS12-02\_EUSK\_1)]
  - Sterilize scissors with alcohol.
  - For larger snakes: Take three ventral scale clips from the largest midbody scales, the three samples not from adjoining scales. The clip should be ~1 mm x ~3 mm, but try to clip all the way across each scale, and try to get some of the pigmentation of each scale.
  - For small snakes and lizards: Snip ~3 mm of the tail tip with scissors into centrifuge tube.

Place drop of tissue adhesive (New Skin) on cut, allow to air dry.

Place micro-centrifuge tube in designated container in specimen freezer at the office.
4. Take photos (Optional except for Mt. Kingsnakes and Rubber Boa)
  - Minimum of 3 (1 dorsal, 1 ventral, 1 close-up of dorsal portion of head).
    - i. Place, in each photo, ruler and tape with date and specimen # (corresponding to order entered on datasheet).
    - ii. Label the photos with photo #s [date, photographer initials, and photo file number (ex. 20091125\_SLP\_362)].
5. Notes - Record unusual morphology
  - Take notes on any unusual characteristics of the animal (e.g. coloration, injuries, regrown tail, etc.).
6. Return animal to exact location where found.

**Non-Target Species Processing Methods (DO NOT PROCESS ANY VENOMOUS REPTILES!)**

1. Gender/Age
  - Male, female or unknown
2. Measurements
  - a. Using metric ruler
    - i. Snout-Vent length (mm)
    - ii. Tail length (mm)
  - b. Using Pesola scale
    - i. Weight (g): tare scale first with bag, then place animal in bag.
      1. Use the smallest scale possible for the most accuracy.
3. Take photos (optional)
  - i. Record photo #s on datasheet.
  - ii. Label the photos with photo #s [date, photographer initials, and photo file number (ex. 20091125\_SLP\_362)].
4. Return animal to exact location where found.

**REFERENCES**

- Richmond JQ, Jockusch EL. 2007. Body size evolution simultaneously creates and collapses species boundaries in a clade of scincid lizards. *Proc. R. Soc. Lond. B.* 274:1701–1708.
- Wood DA, Fisher AN, Reeder TW. 2008. Novel patterns of historical isolation, dispersal, and secondary contact across Baja California in the Rosy Boa (*Lichanura trivirgata*). *Molec. Phylogen. Evol.*:46:484–502.

**Appendix B.** 2010 arroyo toad survey results by stream.

Stream Name	Stream Reach	Arroyo Toad Detected	Date	Assessment or Survey	Wet	Habitat	Slope (%)	Co-occurring Species Detected
Temecula Creek	TMCS40	No	4/28/2010	Assessment	No	good	1	Side-blotched lizard
	TMCS41	No	4/28/2010	Assessment	No	good	0	
Tule Creek (tributary of Temecula Creek)	TULS17	No	6/23/2010	Assessment	No	good	0	California chorus frog, coastal western whiptail, western fence lizard
	TULT3AS4	No	6/23/2010	Assessment	No	good	3	
	TULT3AS5	No	6/23/2010	Assessment	No	good	3	
	TULT3AS6	No	6/23/2010	Assessment	No	good	3	
San Juan Creek	SJUS1	No	6/25/2010	Survey	Yes	poor	3	California newt, bullfrog, California chorus frog, Pacific chorus frog, granite spiny lizard, southern alligator lizard, western fence lizard, speckled rattlesnake, two-striped garter snake
	SJUS2	No	6/25/2010	Survey	Yes	poor	4	
	SJUS3	No	6/25/2010	Survey	Yes	good	6	
	SJUS4	No	6/25/2010	Survey	Yes	good	0	
	SJUS5	No	6/25/2010	Survey	Yes	Moderate	5	
	SJUS6	Yes	6/25/2010	Survey	Yes	Moderate	0	
	SJUS7	Yes	6/28/2010	Survey	Yes	Moderate	2	
	SJUS8	No	6/28/2010	Survey	Yes	good	0	
	SJUS9	No	6/28/2010	Survey	Yes	good	2	
	SJUS10	No	6/28/2010	Survey	Yes	good	1	
	SJUS11	Yes	5/24/2010	Survey	Yes	good	0	
SJUT3S1	No	6/29/2010	Survey	Yes	poor	3		

**Appendix B cont.**

<b>Stream Name</b>	<b>Stream Reach</b>	<b>Arroyo Toad Detected</b>	<b>Date</b>	<b>Assessment or Survey</b>	<b>Wet</b>	<b>Habitat</b>	<b>Slope (%)</b>	<b>Co-occurring Species Detected</b>
	MORS1	Yes	5/24/2010	Survey	Yes	Good	2	
Morrell Creek	MORS2	Yes	7/2/2010	Survey	Yes	good	1	California newt, California chorus frog, Pacific chorus
(tributary of San Juan	MORS3	No	7/2/2010	Survey	Yes	poor	5	frog, granite spiny lizard, western fence lizard, western
Creek)	MORS4	No	7/2/2010	Survey	Yes	poor	8	skink, two-striped garter snake
	MORS5	Yes	7/2/2010	Survey	Yes	Moderate	1	
Long Creek	LCTS1	No	7/2/2010	Survey	Yes	Moderate	5	California newt, California chorus frog, Pacific chorus
(tributary of San Juan	LCTS2	No	7/2/2010	Survey	Yes	Moderate	3	frog, western fence lizard, two-striped garter snake
Creek)	LOSS1	No	5/25/2010	Survey	Yes	good	2	
	LOSS2	No	5/25/2010	Survey	Yes	good	2	
	LOSS3	No	5/25/2010	Survey	Yes	good	1	Black bullhead, bluegill sunfish, unidentified fish,
	LOSS4	No	5/27/2010	Survey	Yes	good	2	bullfrog, California chorus frog, Pacific chorus frog,
Los Alamos	LOSS5	No	6/9/2010	Survey	Yes	good	1	coastal western whiptail, granite spiny lizard, southern
Creek	LOSS6	No	6/9/2010	Survey	Yes	Moderate	1	alligator lizard, western fence lizard, two-striped garter
	LOSS7	No	6/9/2010	Survey	Yes	Moderate	4	snake
	LOSS8	No	6/21/2010	Survey	Yes	good	3	
	LOSS9	No	6/21/2010	Survey	Yes	good	1	
	LOSS10	No	6/21/2010	Survey	Yes	poor	2	

**Appendix B cont.**

<b>Stream Name</b>	<b>Stream Reach</b>	<b>Arroyo Toad Detected</b>	<b>Date</b>	<b>Assessment or Survey</b>	<b>Wet</b>	<b>Habitat</b>	<b>Slope (%)</b>	<b>Co-occurring Species Detected</b>
	SJAS262	No	6/15/2010	Survey	Yes	good	1	
	SJAS263	No	6/15/2010	Survey	Yes	good	2	
	SJAS263	No	6/23/2010	Assessment	Yes	good	2	
	SJAS264	No	6/15/2010	Survey	Yes	good	1	
	SJAS264	No	6/23/2010	Assessment	Yes	good	1	
	SJAS265	No	6/17/2010	Survey	Yes	good	1	Arroyo chub, threespine stickleback, California chorus
San Jacinto River	SJAS266	No	6/17/2010	Survey	Yes	good	2	frog, Pacific chorus frog, western toad, side-blotched
	SJAS271	No	6/8/2010	Survey	Yes	good	1	lizard, western fence lizard, two-striped garter snake
	SJAS271	No	6/23/2010	Survey	Yes	good	1	
	SJAS271	No	7/7/2010	Assessment	Yes	good	1	
	SJAS272	No	6/8/2010	Survey	Yes	good	0	
	SJAS272	No	6/23/2010	Survey	Yes	good	0	
	SJAS272	No	7/7/2010	Assessment	Yes	good	0	
San Jacinto, N.	SJNS16	No	6/2/2010	Survey	Yes	poor	1	California chorus frog, granite spiny lizard, western fence
Fork	SJNS17	No	6/2/2010	Survey	Yes	Moderate	3	lizard, two-striped garter snake
	SJNS18	No	6/2/2010	Survey	Yes	poor	5	

**Appendix B cont.**

<b>Stream Name</b>	<b>Stream Reach</b>	<b>Arroyo Toad Detected</b>	<b>Date</b>	<b>Assessment or Survey</b>	<b>Wet</b>	<b>Habitat</b>	<b>Slope (%)</b>	<b>Co-occurring Species Detected</b>
	SJSS13	No	6/3/2010	Survey	Yes	poor	1	
	SJSS14	No	6/3/2010	Survey	Yes	poor	0	Arroyo chub, threespine stickleback, California chorus
San Jacinto, S.	SJSS21	No	5/28/2010	Survey	Yes	poor	3	frog, granite spiny lizard, side-blotched lizard, southern
Fork	SJSS22	No	5/28/2010	Survey	Yes	poor	2	alligator lizard, western fence lizard, striped racer, two-
	SJSS23	No	7/14/2010	Survey	Yes	poor	4	striped garter snake
	SJSS24	No	7/14/2010	Survey	Yes	poor	2	
	POTS3	No	6/11/2010	Assessment	Yes	good	4	
Potrero Creek	POTS3	No	6/22/2010	Assessment	No	poor	4	
(tributary of San	POTS4	No	6/11/2010	Assessment	Yes	good	0	none
Jacinto River)	POTS4	No	6/22/2010	Survey	Yes	poor	0	
	POTS5	No	6/22/2010	Survey	Yes	poor	10	
	SNDS3	No	6/3/2010	Assessment	No	good	1	
	SNDS4	No	6/3/2010	Assessment	No	good	3	
Sand Creek	SNDS5	No	6/3/2010	Assessment	No	good	2	
(tributary of San	SNDS6	No	6/3/2010	Assessment	No	good	3	none
Jacinto River)	SNDS7	No	6/3/2010	Assessment	No	good	1	
	SNDS8	No	6/3/2010	Assessment	No	good	3	
	SNDS9	No	6/3/2010	Assessment	No	good	2	
	SNDS10	No	6/3/2010	Assessment	Yes	Moderate	6	

**Appendix B cont.**

<b>Stream Name</b>	<b>Stream Reach</b>	<b>Arroyo Toad Detected</b>	<b>Date</b>	<b>Assessment or Survey</b>	<b>Wet</b>	<b>Habitat</b>	<b>Slope (%)</b>	<b>Co-occurring Species Detected</b>
Strawberry Creek (tributary of San Jacinto River)	STRS1	No	5/28/2010	Assessment	Yes	poor	6	
	STRS2	No	5/28/2010	Assessment	Yes	poor	5	
	STRS3	No	5/28/2010	Assessment	Yes	poor	4	none
	STRS4	No	5/28/2010	Assessment	Yes	poor	4	
	STRS5	No	5/28/2010	Assessment	Yes	poor	4	
Toll Road Creek (tributary of San Jacinto River)	TOLS4	No	6/3/2010	Survey	Yes	poor	10	California chorus frog, coastal western whiptail, western fence lizard
	TOLS5	No	6/3/2010	Survey	Yes	poor	6	
Arroyo Seco	ARRS12	No	6/7/2010	Survey	Yes	good	2	
	ARRS13	Yes	6/7/2010	Survey	Yes	good	1	California chorus frog, Pacific chorus frog, orangethroat whiptail, side-blotched lizard, common kingsnake
	ARRS14	Yes	6/7/2010	Survey	Yes	good	0	
	ARRS15	Yes	6/7/2010	Survey	Yes	good	3	
Indian Creek	INDS27	No	6/16/2010	Assessment	Yes	poor	10	
	INDS28	No	6/16/2010	Assessment	Yes	poor	8	
	INDS29	No	6/16/2010	Assessment	Yes	poor	2	
	INDS30	No	6/8/2010	Assessment	Yes	poor	3	
	INDS30	No	6/16/2010	Assessment	Yes	poor	3	California chorus frog
	INDS31	No	6/8/2010	Assessment	Yes	poor	8	
	INDS31	No	7/7/2010	Assessment	Yes	poor	8	
	INDS32	No	7/7/2010	Assessment	Yes	poor	4	



**Appendix B cont.**

<b>Stream Name</b>	<b>Stream Reach</b>	<b>Arroyo Toad Detected</b>	<b>Date</b>	<b>Assessment or Survey</b>	<b>Wet</b>	<b>Habitat</b>	<b>Slope (%)</b>	<b>Co-occurring Species Detected</b>
	MELS1	No	6/16/2010	Survey	Yes	poor	3	
	MELS2	No	6/16/2010	Survey	Yes	poor	5	
	MELS3	No	6/16/2010	Survey	Yes	Moderate	8	
	MELS4	No	6/16/2010	Survey	Yes	poor	8	
	MELS5	No	6/16/2010	Survey	Yes	Moderate	3	
Mellor Creek (tributary of Indian Creek)	MELS6	No	6/8/2010	Assessment	Yes	Moderate	2	California chorus frog, pacific chorus frog, western toad, coastal western fence lizard, granite night lizard, granite spiny lizard, side-blotched lizard, western fence lizard, two-striped garter snake, gopher snake
	MELS6	No	6/24/2010	Survey	Yes	good	2	
	MELS7	No	6/8/2010	Assessment	Yes	Moderate	5	
	MELS7	No	6/24/2010	Survey	Yes	poor	5	
	MELS8	No	6/8/2010	Assessment	Yes	Moderate	4	
	MELS8	No	6/24/2010	Survey	Yes	poor	4	
	MELS9	No	7/7/2010	Assessment	Yes	poor	6	
	MELS10	No	7/7/2010	Assessment	Yes	good	3	
	MELT1S1	No	6/24/2010	Assessment	No	poor	6	

**Appendix B cont.**

<b>Stream Name</b>	<b>Stream Reach</b>	<b>Arroyo Toad Detected</b>	<b>Date</b>	<b>Assessment or Survey</b>	<b>Wet</b>	<b>Habitat</b>	<b>Slope (%)</b>	<b>Co-occurring Species Detected</b>
	BAUS54	Yes	6/10/2010	Survey	Yes	good	1	
	BAUS55	Yes	6/10/2010	Survey	Yes	good	0	
	BAUS56	No	6/10/2010	Assessment	No	good	1	
	BAUS57	No	6/10/2010	Assessment	No	good	2	
	BAUS58	Yes	6/10/2010	Survey	Yes	good	1	
	BAUS59	Yes	6/10/2010	Survey	Yes	good	3	
	BAUS60	No	6/10/2010	Survey	Yes	good	2	
	BAUS61	No	6/10/2010	Survey	Yes	good	0	
Bautista Creek	BAUS62	No	7/1/2010	Assessment	No	good	2	California chorus frog, pacific chorus frog, western toad, coastal western fence lizard, granite spiny lizard, side-blotched lizard, western fence lizard
	BAUS63	No	7/1/2010	Survey	Yes	good	1	
	BAUS64	No	7/1/2010	Survey	Yes	good	1	
	BAUS65	No	7/1/2010	Survey	Yes	good	2	
	BAUS66	No	7/1/2010	Assessment	No	Moderate	3	
	BAUS78	No	7/7/2010	Assessment	No	Moderate	0	
	BAUS79	No	7/7/2010	Assessment	No	Moderate	3	
	BAUS80	No	7/7/2010	Survey	Yes	Moderate	3	
	BAUS81	No	7/7/2010	Survey	Yes	Moderate	1	
	BAUS82	No	7/7/2010	Survey	Yes	poor	3	

**Appendix B cont.**

<b>Stream Name</b>	<b>Stream Reach</b>	<b>Arroyo Toad Detected</b>	<b>Date</b>	<b>Assessment or Survey</b>	<b>Wet</b>	<b>Habitat</b>	<b>Slope (%)</b>	<b>Co-occurring Species Detected</b>
	WILS15	No	6/14/2010	Assessment	Yes	good	1	
	WILS42	No	6/14/2010	Assessment	No	good	1	
	WILS46	No	6/14/2010	Assessment	No	good	0	
	WILS47	No	6/14/2010	Assessment	No	good	2	
	WILS48	No	6/14/2010	Assessment	No	good	1	
Wilson Creek	WILS49	No	6/14/2010	Assessment	No	good	2	Pacific chorus frog
	WILS50	No	6/14/2010	Assessment	Yes	poor	12	
	WILS51	No	6/14/2010	Assessment	Yes	poor	0	
	WILS52	No	6/14/2010	Assessment	No	good	0	
	WILS53	No	6/14/2010	Assessment	No	good	1	
	WILT1S1	No	6/14/2010	Assessment	No	good	0	
	WILT1S2	No	6/14/2010	Assessment	No	good	2	

**Appendix B cont.**

<b>Stream Name</b>	<b>Stream Reach</b>	<b>Arroyo Toad Detected</b>	<b>Date</b>	<b>Assessment or Survey</b>	<b>Wet</b>	<b>Habitat</b>	<b>Slope (%)</b>	<b>Co-occurring Species Detected</b>
	CAHS8	No	6/14/2010	Assessment	Yes	good	3	
	CAHS8	No	6/21/2010	Survey	Yes	Moderate	3	
	CAHS9	No	6/21/2010	Survey	Yes	Moderate	3	
	CAHS10	No	6/21/2010	Assessment	Yes	poor	3	
Cahuilla Creek	CAHS11	No	6/21/2010	Assessment	Yes	poor	5	California chorus frog, pacific chorus frog, western fence lizard
(tributary of Wilson	CAHS12	No	6/21/2010	Assessment	Yes	poor	7	
Creek)	CAHS13	No	6/21/2010	Assessment	Yes	poor	11	
	CAHS14	No	6/21/2010	Assessment	Yes	poor	3	
	CAHS15	No	6/21/2010	Assessment	Yes	poor	2	
	CAHS16	No	6/22/2010	Survey	Yes	good	3	
	CAHS17	No	6/22/2010	Survey	Yes	good	2	